

Prevalence, morphological and biochemical study of larval stage *Coenurus cerebralis* of *Taenia multiceps* in sheep

N.H. Mohammed

Department of Microbiology, College of Veterinary Medicine, University of Mosul, Mosul, Iraq
Email: nadehamed2017@yahoo.com

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Abstract

This study aimed to estimate the prevalence of *Coenurus cerebralis* of *Tenia multiceps* in sheep of Nineveh governorate. One hundred and thirty-six sheep heads were examined for the presence of coenurus cysts 31 (22.8%) were infected. The younger were more affected than the adults. There were no significant differences between the number of infected males and females. The clinical signs varied from one animal to another which consisted of neurological disorders manifested by depression, bending of the head to one side, walking in a circular movement and paralysis in the hind limbs, while some animals did not give the clinical symptoms despite the existence of cyst. The number of cysts were ranged from 1 to 4 cysts/animal. The size of these cyst was ranged from 0.5 to 4 cm, and the volume of fluid in these cysts were ranged from 1 to 6 ml while the number of protoscolices in the cyst were ranged from 60 - 360 protoscolex, the mean length of large and small hooks was 179.95 µm respectively. The study also considered the identification of some chemical constituents of fluid of coenurus cyst such as glucose, total protein, cholesterol, triglycerides, calcium, sodium, potassium, urea and pH.

Keywords: *C. cerebralis*, Sheep, Cyst, *T. multiceps*

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دراسة نسبة حدوثية مع دراسة شكلية وكيمياء حيوية للطور الوسطي *Coenurus cerebralis* للدودة البالغة *Taenia multiceps* في الأغنام

نادية حامد محمد

فرع الأحياء المجهرية، كلية الطب البيطري، جامعة الموصل، الموصل، العراق

الخلاصة

الهدف من الدراسة الحالية هو تشخيص ومعرفة نسبة انتشار الخمج بالأكياس المثانية في الضأن في محافظة نينوى. تضمنت الدراسة فحص ١٣٦ رأس من الضأن ولوحظ وجود الأكياس المثانية في ٣١ رأس وبنسبة خمج ٢٢,٨%. كما لوحظ ارتفاع نسبة الخمج بين الحيوانات الصغيرة مقارنة بالحيوانات الكبيرة وتبين عدم وجود فرق معنوي في نسبة الخمج بين الذكور والإناث. سجلت الدراسة الأعراض السريرية المتمثلة بالاضطرابات العصبية والتي شملت الحركة الدورانية ميلان الرأس الى أحد الجوانب وشلل الأطراف الخلفية وبعض الحيوانات لم يلاحظ عليها أعراض سريرية على الرغم من وجود الأكياس في الدماغ، وتراوح عدد الأكياس المثانية من ١ - ٤ كيس/رأس وتراوح أحجام الأكياس من ٠,٥ - ٤ سم في حين تراوح كمية السائل من ١-٦ مل وبلغ عدد الرؤوس من ٦٠ - ٣٦٠ رأس كما سجل معدل أطوال الشصوص الكبيرة والصغيرة ١٧٩ و ٩٥ ميكرون وعلى التوالي. وشملت الدراسة أيضا الكشف عن بعض المكونات الكيمياءوية لسائل الاكياس المثانية والمتمثلة بقياس سكر الكلوكوز والبروتين الكلي والكوليسترول والدهون الثلاثية والصوديوم والكالسيوم والبوتاسيوم واليوريا وعامل الأس الهيدروجيني.

Introduction

C. cerebralis is one of the most important parasites that are responsible for a high mortality rate up to 100% of sheep herds in different countries of the world (1), causing a serious economic problem affecting sheep breeding programs (2). The cause of this infection is the metacestode *C. cerebralis* of the *T. multiceps* (3). Since the parasite life cycle requires two hosts, dogs are the final host, while the human and the herbaceous animals are an intermediate host (4). The larval stage of this cestode infects the brain of sheep leading to neurological symptoms such as circling, ataxia, isolation from herd and paralysis, as well as blindness, loss of appetite, which in turn lead to reduced animal productivity and death (5). Human infected with parasite if accidentally swallow the egg of this parasite (6). This study aims to give important information on infection with metacestode in Nineveh governorate and number of cysts, sizes with morphometric measurements and some chemical components study in coenurus cysts fluid.

Materials and methods

One hundred and thirty-six sheep were purchased from different butchers of Nineveh governorate with different ages and both sexes. Samples were collected immediately after slaughtering, samples were put in containers and transferred to the Parasitology Laboratory, College of Veterinary Medicine, University of Mosul. Heads was opened by a longitudinal incision using a manual saw, divided into two parts and brains were examined, size and number of the cysts in each brain of infected animals were calculated (7).

Cyst fluid was collected and centrifuged at a rate of 500 cycles for five minutes for the deposition of the protoscolices (8), protoscolices were placed in a petri dish to calculate the number of protoscolices per cyst using magnifying glass, protoscolices taken from the cysts were fixed in 10% buffered formalin and placed on glass slide for morphometric measurements. The supernatant was collected in test tubes for measuring the fluid in each cyst and for biochemical analysis of glucose, total protein, triglycerides, cholesterol, electrolyte (potassium, sodium, calcium) and urea using the analysis kit manufactured by Biolabo and the samples were analyzed by the spectrophotometer and for pH measurement of coenurus cyst fluid (9). Data were analyzed by chi -square test (Jandel sigma stat scientific software V 3.1)

Results

The results revealed the percentage of total infection with coenurus cyst in sheep was 22.8%. The infection rate was high in young sheep compared with that in adult.

Statistical analysis showed significant difference between ages (Table 1), while no significant difference was reported between sexes (Table 2).

The general clinical examination of affected sheep before slaughter showed a number of clinical symptoms varied from one animal to another, which consisted of neurological disorders manifested by depression, bending of the head to one side, circular movement and paralysis in the hind limbs, while some animals did not notice the clinical symptoms despite the existence of cyst.

The results showed that number of *C. cerebralis* observed in the brain ranged from 1 to 4 cyst/animal. The size of these cyst ranged from 0.5 to 4cm. The results indicated that the volume of liquid in these cysts ranged from 1 to 6 ml while the number of protoscolices in the cyst ranged from 60 - 360 protoscolices (Figures 1 and 2).

Table 1: The infection rates of *Coenurus cerebralis* according to animal age

Age (years)	No. animals examined	No. animals infected	% infection
1<	45	14	31.1 ^a
1-2	39	12	30.8 ^a
2-3	25	4	16.0 ^a
≥3	27	1	3.7 ^b

Different letter in column are significantly different at P<0.05.

Table 2: The infection rates of *Coenurus cerebralis* according to animal sex

Sex	No. animals examined	No. animals infected	% infection
Males	82	20	24.4 ^a
Females	54	11	20.4 ^a

Different letter in column are significantly different at P<0.05.



Figure 1: *C. cerebralis* cysts in brain of sheep.



Figure 2: Number of protoscolices appeared as white clusters on the internal layer of the cyst.

The average size of the protoscolices ranged from 0.45 to 0.6 μm the protoscolices have 4 suckers and rostellum hooks, numbered 24-32 it has same number of large and small hooks (Figures 3 and 4). The length of large hooks ranged from 170 to 190 μm while the length of small hooks was 80 to 120 μm (Tables 3 and 4).

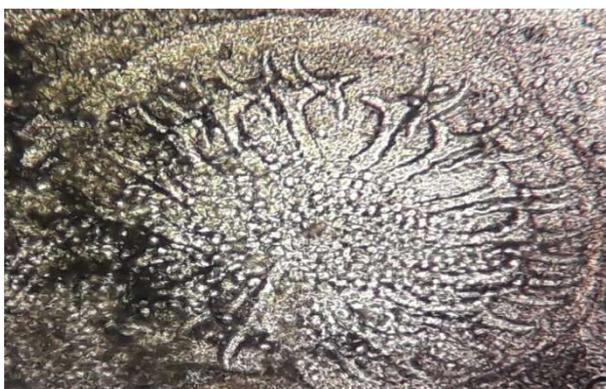


Figure 3: Rostellar hooks with small and large hooks are seen.



Figure 4: The protoscolix has rostellum hooks and suckers.

Table 3: Some of structural characteristics of the *C. cerebralis*

Variables	Range	Mean \pm StE
Number of cysts	1-4	2.3 \pm 0.5
Cyst size (cm)	0.5-4	2.1 \pm 0.3
Fluid volume (ml)	1-6	2.2 \pm 0.6
Number of protoscolices	60-360	149 \pm 1.0
protoscolices size (mm)	0.45-0.6	0.5 \pm 0.2
Number of hooks	24-32	28 \pm 0.3
Large hooks length (μm)	170-190	179 \pm 5.2
Small hooks length (μm)	80-120	95 \pm 0.8

Table 4: Biochemical parameters of *Coenurus cerebralis* cystic fluid

Parameters	Range	Mean \pm StE
Glucose (mg/dl)	130-165	151.6 \pm 20.1
Protein (g/dl)	5-7	6 \pm 0.2
Cholesterol (mg/dl)	130-150	140 \pm 1.0
Triglyceride (mg/dl)	125-150	138 \pm 1.6
Ca (mg/dl)	7.8-10	8.9 \pm 0.9
Na (mmol/l)	85-140	115 \pm 1.8
K (mmol/l)	3.8-5	4.3 \pm 0.5
Urea (mg/dl)	3-4	3.5 \pm 0.2
PH	7-7.5	7.3 \pm 0.1

Discussion

Knowing the percentage of infection with *C. cerebralis* greatly helps in how to prevent and control disease during sheep raising and thus reduce the economic losses caused by it (10). The study explained that the total infection rate was 22.8%, this result is apparently similar to that obtained by Uslu and Guctu (11) who detected coenurus cyst infection in Istanbul of 24.61%, but higher than that carried out in Abog (12) were recorded 2.68%, and lower than the study done in Ethiopian highland and Egypt which were 75 and 100%, respectively (13,14). This difference in the percentage of infection in several studies may have relationship to different efforts in the system of management, breeding, the amount of contamination of pastures with worm eggs and differences in attempts to control the disease (15).

The current study also showed the age of susceptibility to infection and the infection rate of *C. cerebralis* was low in adult compared to that in young sheep. These results agreed with studies carried out on sheep in Iran (16) reported in that young sheep were more susceptible to *C. cerebralis* effects and it is suggested that older sheep eventually develop some kind of immunity to infection (17), on other hand no significant difference in infection rate between males and females this result was in agreement

with Alani and Belli (12). Clinical symptoms of some of the animals were recorded while some of the animals were found to have no clinical symptoms those results were similar to those noticed by Qassim *et al* (18).

In this study, the number of cyst in the animals ranged from 1-4 cyst / infected sheep and this result was higher than the study done by Alani and Belli (12), in that it was found that the number of cyst ranged from 1-2 cyst/ infected sheep but lower than the study in Ethiopian (13), who detected that the number of cyst was ranged from 1-8 cyst/ infected sheep. The measurement protoscolex and hooks observed in this study were in agreement with those described by some authors (19,20).

The results of the study indicated that the concentration of glucose was 151.6 mg/dl, since glucose is a major source of energy for both the metacystode and adult worms, which has a role in the effective transport mechanism (9). the total protein level in the cyst fluid of *C. cerebralis* was 6 g/dL this result is apparently similar to that obtained by Amer *et al* (21) who referred to the concentration of the protein which depends on degree of maturity, the age and history of metabolism of the larval stage of parasite. The concentration of cholesterol and triglyceride in cyst fluid of *C. cerebralis* were 140 mg/dL and 138 mg/dL, respectively. This result was agreement with experiences of Oryan *et al* (9) who reported that cestode tegument and the intermediate phase of the parasite which includes a various type of phospholipids, cholesterol and glycolipids, which the parasite need in order to and survive. The urea in the cyst fluid of the *C. cerebralis* was 3.5 mg/dl, as the presence of urea refers to the occurrence of the ornithine cycle previously detected in *E. granulosus* worms (22). The concentration of K, Na and Ca in the cyst fluid were 4.3 mmol/l, 115 mmol/l and 8.9 mg/dl, respectively. The concentration of minerals in cyst fluid maintaining water and pH balance as the high level of electrolyte inhibits the acidity of the cyst fluid at pH 7.3. These results were in agreement with studies (9,23). These chemical tests are playing an important role in the recognizing of the physiology and immunity and the metabolic role of the intermediate phase of the *Taenia* (24).

Conclusion

From 136 sheep heads were examined for the presence of coenurus cysts 31 (22.8%) were infected, The young animals were more affected than the adults. The number of cysts were ranged from 1 to 4 cysts/animal. The size of these cyst was ranged from 0.5 to 4 cm, and the volume of fluid in these cysts were ranged from 1 to 6 ml while the number of protoscolices in the cyst were ranged from 60 - 360 protoscolex, the mean length of large and small hooks was 179.95 μ m respectively. The study also considered the identification of some chemical constituents of fluid of

coenurus cyst such as glucose, total protein, cholesterol, triglycerides, calcium, sodium, potassium, urea and pH.

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